SEMIOCHEMICALS FOR CAPTURING THE AMBROSIA BEETLE, *TRYPODENDRON LINEATUM*, IN MULTIPLE-FUNNEL TRAPS IN BRITISH COLUMBIA

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Abstract

The host attractants, ethanol and alpha-pinene, and the aggregation pheromone, lineatin, were tested alone and in all combinations for attracting the ambrosia beetle, *Trypodendron lineatum* (Olivier), to Lindgren multiple-funnel traps in a forest setting. A laboratory study examined the flight responses of *T. lineatum* to various release rates of ethanol and lineatin, alone and in combination, in a wind tunnel. Lineatin was the only effective chemical in attracting the beetles to the traps in both studies. There were no synergistic effects from adding ethanol or α-pinene, alone or together, to lineatin-baited traps in the field. The responses of both sexes in the wind tunnel were highest at lineatin release rates of 8 and 64 ug/24 h. A decrease in response occurred at 512 ug/24 h.

INTRODUCTION

The ambrosia beetle, *Trypodendron lineatum* (Olivier), like most species of the family Scolytidae, relies on its olfactory perception of chemicals for host attraction and mating (Borden 1985). Because this insect is a major pest of logged timber on the Pacific coast of British Columbia, a substantial effort has been put into identifying and quantifying the chemicals and the combinations to which the beetle responds. This has resulted in improved methodology in surveying and managing the pest (Borden and McLean 1981; Lindgren *et al.* 1983).

Three chemicals have been identified as attractants for *T. lineatum* in North America; ethanol (Moeck 1970, 1971) and α-pinene (Nijholt and Schonerr 1976) as host attractants, and lineatin, a tri-cyclic ketal (MacConnell *et al.* 1977; Borden *et al.* 1979) as the aggregation pheromone. Combinations of these chemicals, for use in trapping programs, have been investigated in Europe and western North America where *T. lineatum* occurs. Borden *et al.* (1982) confirmed reports from Europe that ethanol, α-pinene, and lineatin acted synergistically in attracting *T. lineatum*. However, they found that ethanol and α-pinene did not enhance the attraction of *T. lineatum* to sticky wire mesh or drainpipe traps in British Columbia. In contrast, Shore and McLean (1983) found that ethanol and α-pinene together did act synergistically with lineatin in attracting *T. lineatum* adults to drainpipe traps.

The different results may be attributed to several factors. Firstly, Borden *et al.* (1982) used release rates of ethanol and α-pinene 3-6 and 2-3 times greater, respectively, than did Shore and McLean (1983). Secondly, Shore and McLean (1983) actually tested for interactions between the semiochemicals of *Gnathotrichus sulcatus* (LeConte) and *T. lineatum*. These interactions may have influenced beetle response to traps baited only with *T. lineatum* semiochemicals. In addition, the latter authors did not differentiate the effects of ethanol and α-pinene in their treatments.

The conflicting results of these studies, along with the development and widespread use of the multiple-funnel trap (Lindgren 1983), prompted us to investigate the importance of these semiochemicals in attracting *T. lineatum* to the efficient new traps. We set out to determine: 1) the optimal combination of semiochemicals needed for attraction (Bedard and Wood 1981; Pearce *et al.* 1975; Renwick 1970) and 2) their optimal release rates (Baker and Linn 1984; Schlyter *et al.* 1987). We designed a field experiment to see which combination of semiochemicals resulted in the highest number of beetles caught, and a wind tunnel study to compare the number of *T. lineatum* caught in traps baited with the pheromone and a host attractant, using various combinations and release rates.

MATERIALS AND METHODS

Experiment 1

An 8 X 8 Latin square design (Steel and Torrie 1960) was used to test the attraction of *T. lineatum* to Lindgren multiple-funnel traps, unbaited and baited with ethanol, α-pinene, and lineatin, alone and in combinations. For each trapping period, which averaged 4 days, traps were assigned new positions randomly so that after eight trapping intervals, all the treatments had been in each of the eight positions once. This was used to control any possible effects of time and position on the trap catches.

The semiochemical release devices used and their placement in the funnel traps were consistent with the procedures used in commercial mass-trapping programs along the B.C. coast at the time of the study[1]. Ethanol (95%) was released from a 40 mL plastic container with a 1 mm-diam-aperture (release rate = 75mg/24 h), placed in the middle funnel of the traps. Alpha-pinene was released from a 4.5 mL glass bottle with a 4 mm-diam-aperture (release rate = 30 mg/24 h), also placed in the middle funnel. A slow-release lineatin lure[2] (release rate = 100 ug/24 h) was set in both the top and bottom funnels of the traps.

The traps were placed about 25 m apart in a forested site near a large log boom storage area, on the North Arm of the Fraser River in Vancouver, B.C. The experiment was run from May 12 - June 12, 1985. The traps were checked daily and every time any of the traps caught about 100 beetles, all the traps were emptied and moved to a new, randomly assigned position. The beetles were counted and sexed for each trapping period of the test.

The data were transformed by $x' = \log_{10}(x+1)$ and analyzed by analysis of variance (ANOVA) and the Student-Newman-Keuls test (SNK)($P \leq 0.05$) using U.B.C. ANOVAR (Greig and Osterlin 1978).

Experiment 2

The attraction of beetles to funnel traps baited with various release rates of ethanol and lineatin, was studied in 1987 in a wind tunnel described by Angerilli and McLean (1984), but since shortened to 3.6 m in length, while the width and height remained at 1.2 m each. The tunnel was fitted with activated charcoal and dust filters. Cool white fluorescent lights (660 W each), situated 2 m above the tunnel, were turned on because *T. lineatum* normally flies during the daylight. The acrylic plastic ceiling of the tunnel was covered with cellulose acetate to minimize glare and diffuse the illumination. Windspeed in the tunnel was maintained at 15 cm/sec, and the temperature averaged 23 ± 2° C.

Three 8-funnel traps were placed in the upwind section of the tunnel, 0.5 m from the screen. The three traps provided a larger plume of semiochemicals within the tunnel than could be achieved with a single trap.

Treatments for testing the reponse of *T. lineatum* in the wind tunnel included 4 lineatin release rates (0, 8, 64, and 512 ug/24 h), each tested in all combinations with 3 release rates of 95% ethanol (0, 75, and 150 mg/24 h). This resulted in 12 combinations of lineatin and ethanol treatments. The 0,0 release rate was the control treatment.

Slow-release lineatin lures were used, in the form of Hercon controlled release dispensers (Kydonieus and Beroza 1981), with release rates based on lure size (7.0 ug/24 h/cm^2)[1]. The lures were aged for one week at ambient temperatures to allow the release rates to stabilize. Six lures were used in each lineatin treatment. A lure was placed in the top and bottom of each trap. The size of each lure used for release rates of 8, 64, and 512 ug/24 h, were 0.19, 1.51, and 12.2 cm^2, respectively.

Ethanol was released from the same device described in experiment 1. For release rate treatments of 75 mg/24h, one lure was placed in the central trap, and for treatments of 150 mg/24 h, one lure was placed in each of the outer traps.

Traps used for the control treatments were not the same as those used for the baited treatments. To see if baited traps carried residue, a comparative test was run to compare beetle response to these traps following bait removal, with the response to the control traps.

The beetles used in this study had been collected during the spring of 1987 with lineatin-baited multiple-funnel traps at the same field site used in experiment 1. Beetles were collected daily and placed in 1 L plastic containers with moistened cloth towels. The containers were stored in a walk-in cooler at 4° C under a 14:10 h (L:D) photoperiod. The containers were checked twice weekly to monitor moisture levels. These beetles survived for more than 100 days.

1 Phero Tech. Inc., 1140 Clark Dr., Vancouver, B.C. V5L 3K3.

2 Biolure Reg. TM Consep Membranes of Bend, Oregon.

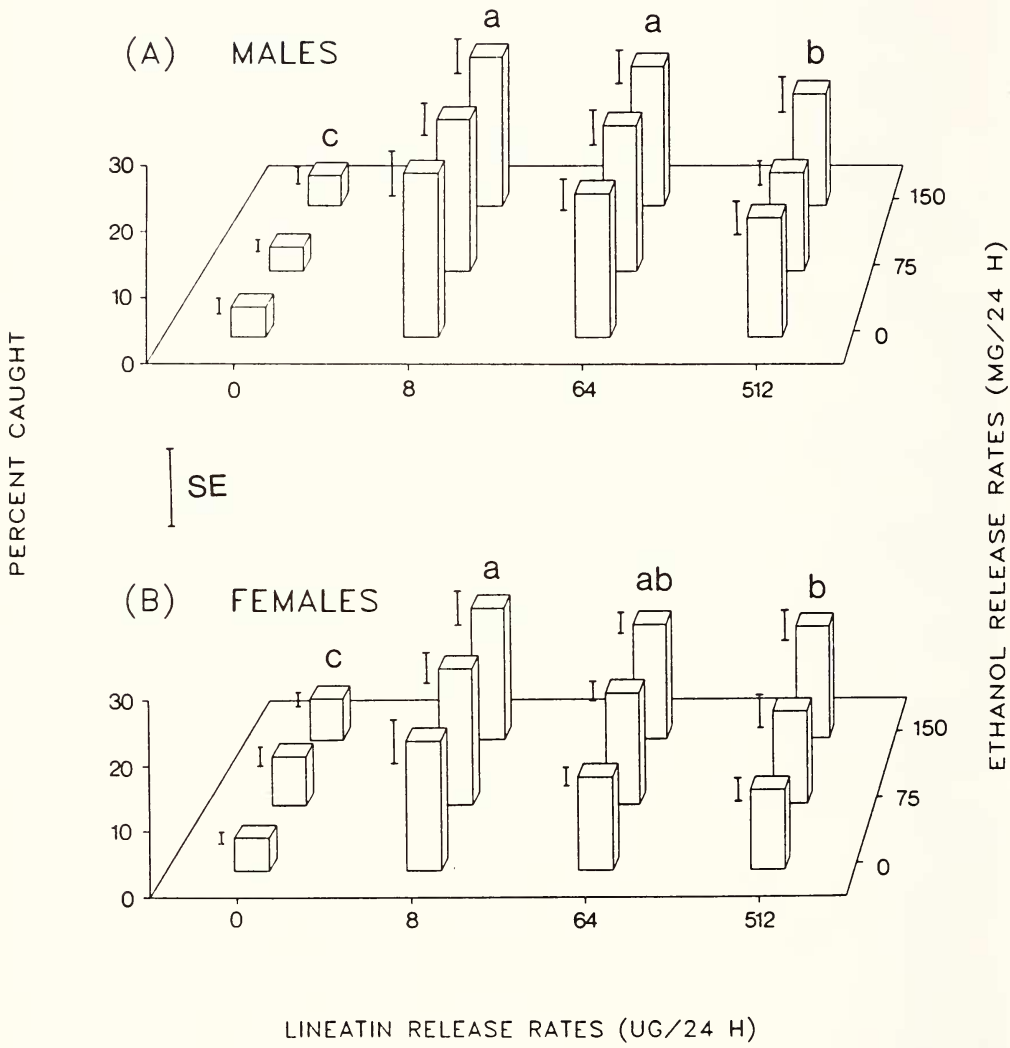

Figure 1. Mean percent (±) SE of *T. lineatum* caught in traps in response to combinations of varied release rates of lineatin and ethanol in a wind tunnel: A) Males and B) Females. Lineatin release rate columns, pooled across ethanol treatments, with the same letter are not significantly different (SNK; $P \leq 0.05$).

Prior to testing, the beetles were removed from the plastic containers and selected for testing based on their healthy appearance (i.e. presence of all body parts) and their ability to walk normally. The selected beetles were then placed in petri dishes with cloth towels and stored at 4° C until needed for the test, always on the same day. The beetles were given a warm-up period of 15 min at room temperature prior to release. The beetles were released from a horizontal tray, 35 cm above the tunnel floor, 1.5 m downwind from the traps.

A 4 X 3 factorial experiment was set up as a randomized complete block design with days serving as blocks. Males and females were tested separately. Because of the large number of treatments, one replication was tested per day for each of 15 experimental days. For each replication, 25 beetles were released and allowed 10 min to respond. The percentage of beetles caught in the traps was used as the dependent variable. The arc sine transformed data were analyzed by ANOVA and mean separation between treatments was carried out with the SNK test ($P \leq 0.05$) (SAS 1985).

RESULTS

Experiment 1

The number of *T. lineatum* caught differed significantly with trapping period ($F_{7,42}$ = 13.3; P kw 0.01), but not with trap position ($F_{7,42}$ = 0.8; P 0.05), demonstrating the importance of the Latin square design in providing confidence that position was not a significant source of variation. Traps baited with lineatin caught significantly more beetles than traps not baited with this chemical (Table 1). Traps baited with ethanol and α-pinene showed no significant differences in catch when compared with the unbaited trap. Ethanol and α-pinene combined with lineatin did not enhance the catch of beetles, and in fact, resulted in a slightly lower catch than the treatment with lineatin alone.

Experiment 2

The number of *T. lineatum* caught in funnel traps in the wind tunnel was not significantly affected by the presence of ethanol either for males ($F_{2,154}$ = 0.4; P 0.05) or females ($F_{2,154}$ = 1.4; P 0.05). In contrast, a significant increase in the number of beetles caught did occur with the presence of lineatin for both males ($F_{3,154}$ = 61.9; $P < 0.01$) and females ($F_{3,154}$ = 32.0; $P < 0.01$).

Males responded best at the lowest release rates of lineatin with an average trap catch of 23.5 and 21.6% for the 8 and 64 ug/24 h release rates, respectively (Figure 1A). A significant reduction in response occurred at 512 ug/24 h with a mean catch of 16.6% of the beetles tested. Despite somewhat lower catches, similar responses were found for females (Figure 1B), in which the highest catches, 20.0 and 16.1%, were obtained at 8 and 64 ug/24 h, respectively. Mean catch at 512 ug/24 h was 14.3 %, a significantly lower response than for the 8 ug treatment.

Table 1. Response by *Trypodendron lineatum* to semiochemical-baited funnel traps set in a forest from May - June, 1985.

	Mean Catch/Trap/ Sampling Period		
Bait	Males	Females	Total
Control (unbaited)	0.3 b[1]	0.1 b	0.4 b
Ethanol (E)	2.3 b	3.4 b	5.7 b
Alpha-pinene (P)	0.8 b	0.5 b	1.3 b
E + P	0.4 b	0.6 b	1.0 b
Lineatin (L)	735.0 a	616.0 a	1351.0 a
L + E	415.9 a	438.5 a	854.4 a
L + P	590.1 a	328.6 a	918.7 a
L + E + P	510.9 a	466.8 a	977.7 a

1 Means followed by the same letter not significantly different (SNK; $P \leq 0.05$).

No significant differences in response by *T. lineatum* were observed between the control traps and unbaited traps that had previously held ethanol and lineatin during the experiment ($F_{5,72}$ = 0.9; P 0.05). Thus, there was no evidence for contamination of the traps.

DISCUSSION

The hypothesis supporting the use of ethanol in trapping ambrosia beetles is that it acts as an arrestant/boring stimulant for beetles in close proximity to a suitable host (McLean and Borden 1977). Drainpipe traps require that beetles land on the trap and crawl through small diam holes in order to enter the trap (Bakke 1983). This contrasts with the immediate knockdown capture of beetles in funnel traps (Lindgren 1983). Our data suggest that there is no gain in adding ethanol and/or α-pinene baits to funnel traps, as the lineatin alone is sufficient to attract *T. lineatum* close enough to the trap for capture.

It is possible that ethanol and α-pinene may be important in attracting *T. lineatum* to funnel traps, but that the dispensers normally used for releasing them may account for our results. This is being investigated (Cushon unpublished)[3]. Nevertheless, the same dispensers were shown to be effective in enhancing the attraction of *T. lineatum* in Europe as well as another ambrosia beetle species, *Gnathotrichus sulcatus* (LeConte) to sticky wire mesh traps (Borden *et al.* 1982).

Response of *T. lineatum* to various release rates of lineatin has been previously examined in a field setting in British Columbia by Lindgren *et al.* (1983). They found that the number of beetles caught on cylindrical traps baited with lineatin, increased as release rates increased, from 10-40 ug/24h, and remained the same between 40 and 800 ug/24h. In a later experiment from the same paper, release rates of 40 ug/24h were found to be optimal for funnel traps, yet a release rate of only 10 ug/24 h of lineatin was adequate as long as the remaining lineatin (30 ug/24 h) was placed within 1.5 - 2 m of the trap. Our results in the wind tunnel with low release rates correspond well with those from Lindgren *et al.* (1983). However, a decrease in response was observed in the wind tunnel at the higher release rates. The differences in sensitivity of the beetles at the higher rates may have resulted from artificial factors imposed by the wind tunnel, such as an enclosed environment and a constant, unidirectional air flow. Both factors resulted in continuous exposure of the beetles to the pheromone, whereas in the field, pheromone plumes are often broken up by turbulence and wind shifts (Fares *et al.* 1980), resulting in non-continuous exposure. For these reasons and because the lures weaken with age, it is probably best for mass-trapping in the field to keep the release rates of lineatin higher than for studying beetle flight behavior in the wind tunnel.

Host volatiles may well be important in initial host recognition by *T. lineatum* in a natural forest situation. However, from our results, lineatin baits alone appear to be sufficient for running an effective mass-trapping program with Lindgren funnel traps around log booms and dryland sorts, where host volatiles are likely to be present anyway. The expense and extra labor involved in maintaining the ethanol and α-pinene baits would not seem to be necessary for maintaining effective mass-trapping programs for *T. lineatum* in British Columbia.

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